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Effects of Sevin on Development of Experimental Estuarine Communities

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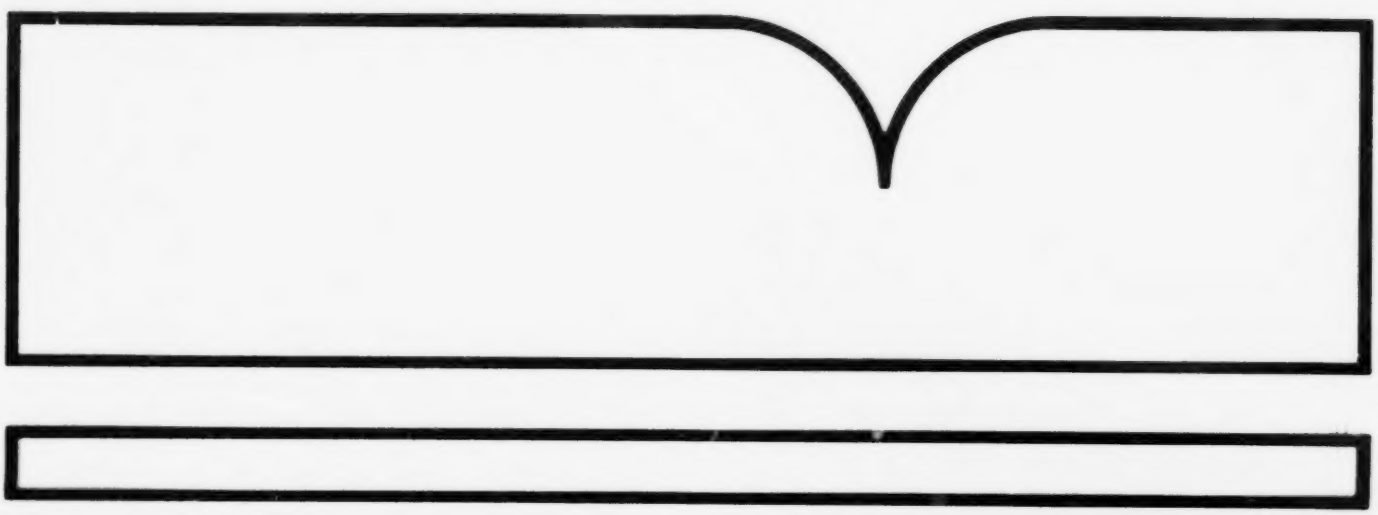

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16. ABSTRACT

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EFFECTS OF SEVIN ON DEVELOPMENT OF EXPERIMENTAL ESTUARINE COMMUNITIES

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The composition of animal communities developing from planktonic larvae in aquariums containing sand and flowing estuarine water was altered in the presence of the carbamate insecticide Sevin (carbaryl). Treatments were control and concentrations of Sevin that averaged 1.1, 11.1, and 103 μg/l; each treatment was replicated 8 times. Animals that colonized aquarium sand were collected in a 1-mm mesh sieve after 10 wk of exposure. Mollusks, arthropods, annelids, and nemerteans were the numerically dominant phyla. The average number of species per aquarium was significantly less ($\alpha = 0.05$) in aquariums containing 11.1 or 103 μg/l than in those containing 1.1 μg/l or in control aquariums. The abundant clam Ensis minor *grew significantly less in length at the higher concentrations of Sevin. The amphipod* Corophium acherusicum *was particularly affected; significantly fewer were found at all concentrations than in the control aquariums. A marked increase in the abundance of the annelid* Polydora ligni *in aquariums containing 103 μg/l corresponded to a marked decrease in the number of other annelids and to a significant absence of nemerteans.*

INTRODUCTION

Sevin (1-naphthyl *N*-methylcarbamate), a synthetic carbamate insecticide also known as carbaryl, has been used in the United States for more than 20 yr. Widespread usage to control agricultural, forestry, poultry, and animal pests in recent years has increased its potential for entering and affecting the estuarine environment.

A variety of estuarine organisms have been experimentally exposed to Sevin. It has been tested in the laboratory (Andrews et al., 1968) and in

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the field (Lindsay, 1961; Haven et al., 1966) for control of pests and predators of oysters such as oyster drills *Ocinebra japonica* and *Urosalpinx cinerea*, ghost shrimp *Callianassa* sp., and pea crabs *Pinnotheres ostreum*. Pea crabs were killed when the oyster hosts were exposed to 10 ppm (mg/l) in the laboratory. In field tests, Sevin with other compounds eliminated ghost shrimp from oyster areas but had little effect on oyster drills. Although the concentrations of Sevin used in these field applications were not given, Karinen et al. (1967) reported that it has been sprayed at rates up to 10 lb/acre (11.2 kg/ha) in field use against oyster pests.

In laboratory experiments, 1 ppm Sevin significantly reduced feeding activity of the lugworm *Arenicola cristata* (N. I. Rubinstein, personal communication).

Sevin was shown to be toxic to marine mollusks at concentrations ranging from 1.0 to 7.3 ppm. Acute toxicity (24-, 48-, or 96-h exposures) for bay mussel *Mytilus edulis*, Pacific oyster *Crassostrea gigas*, and cockle clam *Clinocardium nuttalli* ranged from 2.3 to 7.3 ppm (Stewart et al., 1967; Butler et al., 1968). The toxicity of the hydrolytic product of Sevin, 1-naphthol, to mollusks was approximately the same as that of the parent compound. Development of clam embryos (*Mercenaria virginica*) was inhibited by 2.5 ppm Sevin, and oyster embryos (*Crassostrea virginica*) by 1 ppm (Davis, 1961).

Marine arthropods, particularly shrimp, are relatively sensitive to Sevin at concentrations ranging from 2.5 to 1000 ppb (μg/l). Concentrations (ppb) causing acute effects (loss of equilibrium, paralysis, or death in 24-, 48-, or 96-h exposures) were 1000 for stone crab *Menippe mercenaria*; 600 for Dungeness crab *Cancer magister*; 550 for blue crab *Callinectes sapidus*; 400 for mud shrimp *Upogebia pugettensis*; 270 for shore crab *Hemigrapsus oregonesis*; 130 for ghost shrimp *Callianassa californiensis*; 38 for grass shrimp *Palaemonetes pugio*; 7 for Korean shrimp *Palaemon macrodactylus*; and 2.5 for brown shrimp *Penaeus aztecus* (Butler, 1962, 1963; Stewart et al., 1967; Earnest, 1971; Hansen et al., 1973). The 96-h LC50 for the estuarine mysid *Mysidopsis bahia* exposed to carbaryl was 7.7 ppb (DelWayne Nimmo, personal communication). A concentration as low as 0.1 ppb delayed molting in zoeae of the Dungeness crab *C. magister* (Buchanan et al., 1970). Stewart et al. (1967) found that Sevin was more toxic to mud shrimp, ghost shrimp, shore crabs, and Dungeness crabs than was its hydrolytic product 1-naphthol..

Studies indicate that the toxicity of Sevin to fish is about equal to its toxicity to mollusks but less than its toxicity to arthropods. Juvenile spot *Leiostomus xanthurus* survived 5 mo of continuous exposure to 0.1 ppm (Lowe, 1967). The 96-h LC50 for juvenile striped bass *Morone saxatilis* was more than 1 ppm (Korn and Earnest, 1974); the 48-h LC50s for longnose killifish *Fundulus similis* and white mullet *Mugil curema* were 1.75 and 2.5 ppm, respectively (Butler, 1963). The 24-h EC50s (paralysis or death) reported by Stewart et al. (1967) for shiner perch *Cymatogaster*

aggregata, English sole *Parophrys vetulus*, and stickleback *Gasterosteus aculeatus* exposed to Sevin ranged from 3.9 to 6.7 ppm; 1-naphthol was more toxic (EC50s from 1.3 to 3.2 ppm). Weis and Weis (1975) found that as little as 10 ppb Sevin retarded regeneration of caudal fins in the killifish *Fundulus heteroclitus*.

Sevin was also found to be toxic to phytoplankton. Butler (1962) reported that 5 kinds of phytoplankton utilized by molluscan larvae as food could not tolerate Sevin at concentrations higher than 0.1 ppm. Ukeles (1962) found that 1 ppm completely suppressed growth of 2 species of marine algae; 10 ppm was lethal to 3 species.

Since Sevin is known to be toxic to various aquatic organisms, it is likely to affect the development of an estuarine community as a whole. The present study was designed to determine the effects of Sevin on estuarine community development, toxic levels, sensitive species, and growth effects.

METHODS

The effect of Sevin on colonization was determined by comparing numbers and species of animals that grew from planktonic larvae in treated aquariums for 10 wk (January 9 to March 20, 1978) with those of animals that grew in untreated aquariums. Larvae entered the aquariums from the natural component of plankton in flowing unfiltered seawater. We used 4 plastic apparatuses (Fig. 1), each consisting of a central constant-head box and 8 aquariums (40 cm long, 10 cm wide, and 12 cm high). The design of our system was based on work by Hansen (1974), who found that small replicate aquariums readily indicated the effect of a toxicant on developing estuarine animal communities in sand. Aquariums were filled to a depth of 5 cm with clean silica sand (particle sizes: 75%, 354–500 μm; 96%, 250–710 μm) dredged 1 yr previously from Santa Rosa Sound, Florida. Water levels in aquariums were maintained at 8 cm (3 cm above substratum).

Seawater with its constituent plankton was pumped from Santa Rosa Sound to a splitter box where 4 adjacent glass tubes supplied water at a rate of 2 l/min to the 4 constant-head boxes that supplied water to the aquariums (Fig. 1). Flow to each aquarium was maintained at 200 ml/min by adjusting the height of a 3-mm diameter hole (delivery port) in the wall of the constant-head box. (Holes were in movable slides that covered larger holes in the box walls.) Water flowed from each aquarium through a notched end-opening; large predators (e.g., crabs wider than 10 mm) escaped through these openings before their numbers could drastically affect community structure. Salinity of the water averaged 20 ppt (9–30 ppt); temperature, 10°C (5–16°C).

Technical grade Sevin (99.5%) dissolved in triethylene glycol (TEG) was metered by syringe pump into and mixed with the seawater entering

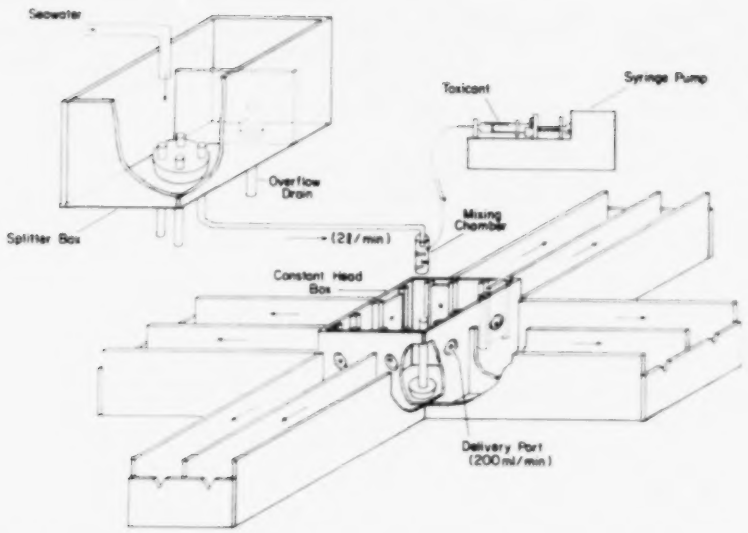

FIGURE 1. Apparatus used to test the effect of Sevin on development of estuarine macrobenthic communities.

the center of the constant-head box of each treated apparatus. The same amount of TEG (10 ml/d, 3.5 μg/l) was metered into the control apparatus. Desired concentrations of Sevin in seawater were 1, 10, and 100 μg/l (ppb).

Samples of water were taken from constant-head boxes twice a week and sand cores were removed from three of eight aquariums from each apparatus at the end of the exposure for analyses for Sevin. The toxicant was extracted from seawater and air-dried sand with methylene chloride and concentrated on a steam table. Silica gel cleanup was used for the sand extracts.

Sevin concentrations were determined with a Waters model 204 high-pressure liquid chromatograph equipped with a μNH_2 Bondapak column. Operating conditions were: carrier, 10% (v/v) isopropanol isooctane; flow rate, 2 ml/min; detector, 280-nm wavelength at 0.01 absorbance units full scale. Average recovery exceeded 90% for seawater and 80% for sand; data were not adjusted for recovery. All samples were fortified with the internal standard carbofuran before analysis to evaluate the integrity of the results. Limits of detection were 1.0 μg/l in seawater and 500 μg/kg (dry weight) in sand (for undetectable amounts, < 1.0 μg/l, we used 0.5 μg/l to calculate average concentration in water). Levels of the metabolite 1-naphthol were not determined.

After 10 wk, animals were collected by siphoning the contents of the aquariums into a 1-mm mesh sieve, after which they were preserved and

identified. Analysis of variance and Duncan's multiple range test were used to compare numbers and species of animals in control and treated aquariums ($\alpha = 0.05$). Results are presented as pooled data for each Sevin concentration and the control.

Lengths of 150 jackknife clams *Ensis minor* were measured for the control and for each concentration to determine whether Sevin affected growth. All jackknife clams collected from each apparatus were dumped into a tray marked with square divisions and measured on a selected sequence of squares (the sequence for all apparatuses began with the same corner square and progressed to adjoining squares until 150 clams were measured). Growth measurements were compared by analysis of variance and Duncan's multiple range test ($\alpha = 0.05$).

RESULTS AND DISCUSSION

The system design readily allowed the determination of effects of Sevin on developing macrofauna. In the analysis of the data by phylum, differences between replicates were not statistically significant, indicating that interaquarium variability was not great and that the eight aquariums for each treatment represented true replicates.

The averages and ranges of Sevin concentrations measured in water from the three treated apparatuses were 1.1 μg/l (< 1.0-2.4), 11.1 μg/l (2.6–28), and 103 μg/l (78–119). None was found in the control aquariums. Sediment samples from the control and treated aquariums had no detectable Sevin.

Our harvest at 8 wk was 7844 animals, representing 29 species of 7 phyla (Table 1). The three most abundant species were the clam *E. minor* (5728), the barnacle *Balanus improvisus* (1300), and the polychaete worm *Polydora ligni* (333). Numerically dominant phyla were Mollusca, Arthropoda, Annelida, and Nemertea (Table 2).

Development of animal communities was altered in the presence of Sevin (Table 3). Numbers of mollusks decreased, although not significantly, with increasing concentration. The growth in length of the abundant clam *E. minor* (size range, 3–20 mm) was significantly less in aquariums that contained 11.1 or 103 μg/l than in aquariums that contained 1.1 μg/l or in control aquariums. Average lengths (mm) were: 8.5 (control), 8.0 (1.1 μg/l), 6.7 (11.1 μg/l), and 6.4 (103 μg/l).

Arthropods were less abundant and the average number of species per aquarium was significantly less in aquariums treated with 11.1 or 103 μg/l than in aquariums treated with 1.1 μg/l or in control aquariums. The amphipod *Corophium acherusicum* was particularly sensitive to Sevin. Abundance of this species significantly decreased at 1.1 μg/l, and it was absent at higher concentrations.

Sevin may have caused changes in biological interactions that affected the relative abundance of species. A significant increase in the abundance

TABLE 1. Animals Collected from Control Aquariums and from Aquariums Exposed to Sevin for 10 wk

Animal	Control	Sevin (μg/l)			Total no. of animals
		1.1	11.1	103	
Mollusca					
Ensis minor	1626	1507	1315	1280	5728
Abra aequalis	52	49	21	35	157
Polinices lacteus	13	7	4	4	28
Acteocina canaliculata	0	0	0	1	1
Cerithidea costata	0	0	0	1	1
Arthropoda					
Balanus improvisus	336	367	332	265	1300
Corophium acherusicum	31	15	0	0	46
Anoplodactylus lentus	5	8	4	4	21
Pagurus longicarpus	4	3	0	0	7
Callinectes sapidus	2	2	0	0	4
Cyclaspis varians	0	2	0	0	2
Oxyurostylis smithi	0	1	0	0	1
Pinnixa chaetopterana	1	0	0	0	1
Porcellanidae	1	0	0	0	1
Praunus flexuosus	0	1	0	0	1
Annelida					
Polydora ligni	43	44	57	189	333
Spiophanes bombyx	50	41	20	6	117
Neanthes succinea	1	2	1	2	6
Nereis pelagica	2	4	0	0	6
Eumida sanguinea	2	1	0	2	5
Capitella capitata	2	1	0	0	3
Armandia agilis	0	1	0	1	2
Ampharete sp.	1	0	0	0	1
Anaitides erythrophyllus	0	0	1	0	1
Chone duneri	1	0	0	0	1
Nemertea					
Nemertea	16	23	20	0	59
Coelenterata					
Actiniidae	3	5	0	0	8
Platyhelminthes					
Coronodena mutabilis	0	2	0	0	2
Echinodermata					
Leptosynapta inhaerens	0	0	1	0	1

of the annelid *P. ligni* in aquariums that contained 103 μg/l corresponded to a significant decrease in the number of other annelids and to a significant absence of nemerteans. Also, the average number of annelid species per aquarium was significantly less at the highest concentration than at the lowest concentration or the control. The high abundance of *P. ligni* in aquariums treated with 103 μg/l may have resulted from lessened competititon for food and space with another spionid, *Spiophanes*

TABLE 2. Animals and Species, by Phylum, Collected from Control Aquariums and Aquariums Exposed to Sevin for 10 wk

			Sevin (μg/l)					
	Control		1.1		11.1		103	
Phylum	No.	Species	No.	Species	No.	Species	No.	Species
Mollusca	1691	3	1563	3	1340	3	1321	5
Arthropoda	380	7	399	8	336	2	269	2
Annelida	102	8	94	7	79	4	200	5
Nemertea	16	1	23	1	20	1	0	0
Coelenterata	3	1	5	1	0	0	0	0
Platyhelminthes	0	0	2	1	0	0	0	0
Echinodermata	0	0	0	0	1	1	0	0
All phyla	2192	20	2086	21	1776	11	1790	12

bombyx, whose numbers progressively decreased with increased concentrations of Sevin. *P. ligni* has a great reproductive capacity. Galtsoff (1964) reported that reproduction of this species on oyster bottoms of Delaware Bay was occasionally so rapid that a deposit of mud several inches thick, consisting of innumerable living worms and their tubes, covered the affected area.

TABLE 3. Average Numbers of Animals and Species per Aquarium, by Phylum, Collected from Control Aquariums and Aquariums Exposed to Sevin for 10 wk

			Sevin (μg/l)					
	Control		1.1		11.1		103	
Phylum	No.	Species	No.	Species	No.	Species	No.	Species
Mollusca	211.4 (154–273)[a]	3.0 (3)	195.4 (172–213)	2.5 (2–3)	167.5 (92–264)	2.2[b] (2–3)	165.1 (122–244)	2.5 (1–3)
Arthropoda	47.5 (29–61)	3.2 (1–6)	49.9 (32–74)	3.4 (2–5)	42.0 (21–67)	1.4[b] (1–2)	33.6 (11–60)	1.4[b] (1–2)
Annelida	12.8 (2–19)	2.8 (2–4)	11.8 (6–19)	3.0 (2–4)	9.9 (2–23)	2.0 (1–3)	25.0 (1–53)	1.8[b] (1–3)
Nemertea	2.0 (0–5)	0.8 (0–1)	2.9 (0–5)	0.9 (0–1)	2.5 (0–5)	0.8 (0–1)	0[b] (0)	0[b] (0)
Other phyla	0.4 (0–3)	0.1 (0–1)	0.9 (0–3)	0.6 (0–2)	0.1 (0–1)	0.1 (0–1)	0 (0)	0 (0)
All phyla	274.0 (198–356)	9.9 (8–14)	260.8 (238–286)	10.4 (8–12)	222.0 (126–333)	6.5[b] (5–8)	223.8 (167–324)	5.6[b] (4–7)

[a] Range is given in parentheses.
[b] Significantly less than control at the 5% level.

The absence of nemerteans at the highest concentration may have been caused not only by direct toxicity of Sevin but also by competition for available sediment with the large number of tube-building *P. ligni*. Field studies of a mud flat in Mitchell Bay, Washington, by Woodin (1974) revealed competition for surface space between burrowing and tube-building forms of polychaete worms; abundances of the two forms were highly significantly correlated.

The presence of Sevin in estuarine waters at the concentrations tested in the laboratory could affect colonization of various mollusks, arthropods, annelids, and nemerteans. Karinen et al. (1967) found that within about 5 d, juvenile clams, nemerteans, and polychaetes surfaced on the mud and became paralyzed, whereas all mud shrimp and amphipods died in aquariums containing bay mud with 5–25 mg of Sevin per liter of water. In our study, 11.1 and 103 μg/l of the toxicant adversely affected the number of individuals and particularly the number of species in developing estuarine communities of animals: 11 and 12 species were collected from aquariums containing 11.1 and 103 μg/l, whereas 20 and 21 species were collected from the control and 1.1 μg/l aquariums (Table 2). In addition, the average number of species per aquarium was significantly smaller at 11.1 and 103 μg/l (Table 3). In a natural community developing under stress by Sevin, adverse effects on species composition would also lessen the community's usefulness as a food source to other organisms in estuarine food webs.

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